ECOLOGY

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Synergistic roles of climate warming and human occupation in Patagonian megafaunal extinctions during the Last Deglaciation

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The causes of Late Pleistocene megafaunal extinctions (60,000 to 11,650 years ago, hereafter 60 to 11.65 ka) remain contentious, with major phases coinciding with both human arrival and climate change around the world. The Americas provide a unique opportunity to disentangle these factors as human colonization took place over a narrow time frame (~15 to 14.6 ka) but during contrasting temperature trends across each continent. Unfortunately, limited data sets in South America have so far precluded detailed comparison. We analyze genetic and radiocarbon data from 89 and 71 Patagonian megafaunal bones, respectively, more than doubling the high-quality Pleistocene megafaunal radiocarbon data sets from the region. We identify a narrow megafaunal extinction phase 12,280 ± 110 years ago, some 1 to 3 thousand years after initial human presence in the area. Although humans arrived immediately prior to a cold phase, the Antarctic Cold Reversal stadial, megafaunal extinctions did not occur until the stadial finished and the subsequent warming phase commenced some 1 to 3 thousand years later. The increased resolution provided by the Patagonian material reveals that the sequence of climate and extinction events in North and South America were temporally inverted, but in both cases, megafaunal extinctions did not occur until human presence and climate warming coincided. Overall, metapopulation processes involving subpopulation connectivity on a continental scale appear to have been critical for megafaunal species survival of both climate change and human impacts.

INTRODUCTION

The loss of Late Pleistocene megafaunal diversity in South America was among the greatest of any continent (52 genera, 83%), and Fell's Cave in Patagonia was one of the first sites globally where archaeological evidence of human hunting was associated with megafaunal remains (*1*–*3*). Despite this, the timing and nature of the South American megafaunal extinctions and the evolutionary relationships of many taxa remain poorly understood. Patagonia provides an ideal study area in this respect because of the excellent preservation of Late Pleistocene fossil remains compared to the rest of South America (*4*, *5*), and the broadly synchronous but antiphase climate changes to those in the Northern Hemisphere (commonly referred to as the bipolar seesaw) (*6*, *7*).

Following maximum glacial extent across the region 28.5 thousand years ago (ka) (*8*), long-term warming from 18 ka to the Holocene was interrupted between ~14.4 and 12.7 ka by a marked 1700-year decline in temperature and glacial advance identified as the Antarctic Cold Reversal (ACR) stadial (Fig. 1) (*7*, *9*). Glacial conditions persisted until ~12.6 ka, when rapid warming and retreat of the Patagonian Ice Sheet led to the expansion of *Nothofagus* forest ~12.3 ka, with peak Holocene warming attained by 11.4 ka (*10*, *11*). Crucially, the first records of human presence in southern Chile occur immediately prior to the ACR stadial, at Monte Verde near Puerto Montt on the western edge of Patagonia, before ~14.6 ka (*12*, *13*). The central plateau of northern Patagonia was occupied before ~13.2 ka (*14*), with the first confirmed occupation in southern Patagonia between ~13.2 and 12.9 ka, and ~12.7 ka in Tierra del Fuego (*14*, *15*). In contrast to the relatively intense occupation of the central plateau, the first human occupations in southern Patagonia appear relatively ephemeral, with the number of sites and intensity of occupation increasing markedly from the early Holocene (*15*, *16*).

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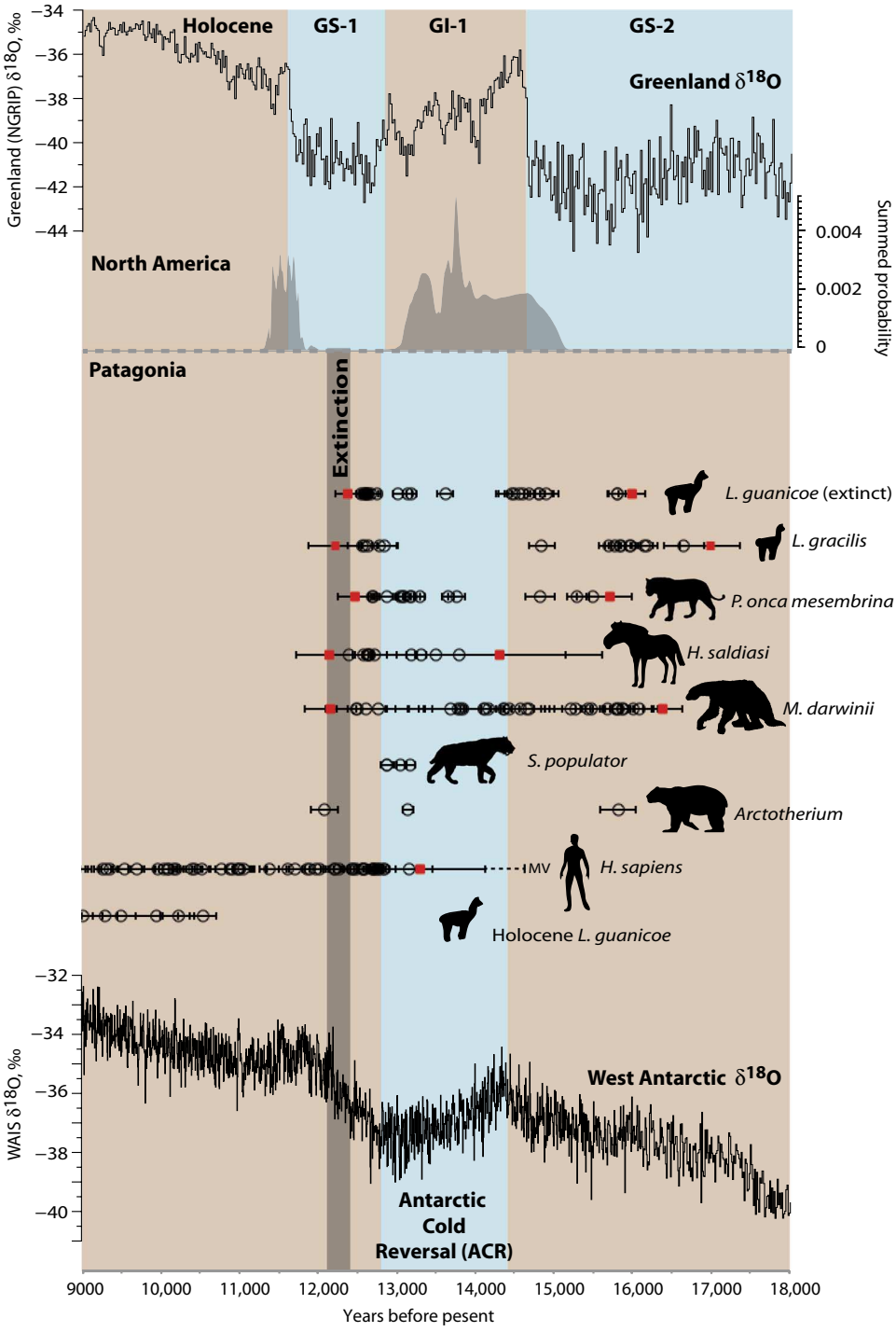

Fig. 1. Extinction chronology of Pleistocene megafauna in southern Patagonia (9000 to 18,000 years ago), shown against West Antarctic Ice Sheet Divide ice $\delta^{18}O$ record (bottom) and the timing of Antarctic Cold Reversal chronozone (blue column) (*9*). Calibrated radiocarbon (^{14}C) ages of extinct megafaunal remains from Patagonia [open circles with 1σ uncertainty indicated by vertical whiskers] are shown with calculated Phase boundary start and end (extinction) estimates (filled red squares) using the Southern Hemisphere calibration (SHCal13) curve (*51*) and OxCal 4.2 (*26*). The vertical gray bar denotes the 1σ range for megafaunal extinction in Patagonia. Calibrated ^{14}C ages for the arrival of humans (with Monte Verde shown as "MV" with dashed line for extended age range) and modern lineage of *L. guanicoe* are shown as open circles and red square. (**Top**) A summary of the Northern Hemisphere climate and North American megafaunal extinctions is also presented. The Greenland $\delta^{18}O$ and isotope event stratigraphy are shown (GS, Greenland Stadial; GI, Greenland Interstadial) (*60*), with the summed probabilities (1σ) of the youngest ^{14}C dates for the latest Pleistocene megafaunal extinctions in North America (*27*), using the IntCal13 calibration curve (*59*). The tan and blue shading highlights the inverted timing of warm and cold intervals in the Northern Hemisphere and Southern Hemisphere during the Last Termination. WAIS, West Antarctic Ice Sheet; NGRIP, North Greenland Ice-Core Project.

To characterize the extinction of the Late Pleistocene Patagonian megafauna, we sequenced mitochondrial DNA (mtDNA) from 89 megafaunal bone and teeth samples recovered from caves and rock shelters, and generated new accelerator mass spectrometry radiocarbon (AMS ^{14}C) dates for a total of 71 bone, teeth, and coprolite samples (tables S1 to S3). We then investigated whether the megafaunal extinctions were associated with major events in records of climate change and/or human occupation.

RESULTS

Phylogenetic relationships of Pleistocene megafauna

Phylogenetic analysis of the new genetic data (see the Supplementary Materials) reveals that the Late Pleistocene megafaunal extinctions contained several previously unknown events, including a distinct camelid species, *Lama gracilis* (*17*), a previously unknown genetic clade of guanaco (*Lama guanicoe*) (Fig. 2), and a genetically distinct giant South American jaguar subspecies, *Panthera onca mesembrina* (*18*). In contrast, the single mitochondrial haplotype recovered from ancient southern Patagonian puma specimens (*Puma concolor*) appears closely related to haplotypes surviving in South America at present.

Extinction of Pleistocene megafauna and associations with human occupation and climate

We combined the newly dated specimens with previously published ^{14}C data of extinct megafauna, such as the large ground sloth (*Mylodon darwinii*) (*19*, *20*), the saber-toothed cat (*Smilodon populator*) (*21*, *22*), the giant short-faced bear (*Arctotherium* sp.) (*23*), and the South American horse (*Hippidion saldiasi*) (*24*, *25*), to compare extinction dates with the arrival of both humans and the genetically distinct modern lineage of *L. guanicoe* (Fig. 1). We used the Phase calibration function with Outlier analysis in OxCal 4.2 to define the midpoint of the start and end (estimated extinction) of the set of ages for each species (*26*), which revealed that the megafaunal extinctions most likely occurred in a narrow time window spanning 12.2 to 12.5 thousand years (table S4). Individual age calibrations of the youngest samples confirm that extinction took place around this time (table S4). Using the C_Combine function in OxCal, we find that the ages for the different species are statistically indistinguishable, implying that extinction across Patagonia was synchronous, with a mean weighted age of 12,280 ± 110 years ago. By doubling the number of high-quality ^{14}C-dated megafaunal fossils for this region, we do not observe carnivore extinctions preceding that of herbivores as recently suggested (*5*). Humans were clearly present in Patagonia for at least 1000 years before the extinction event (Fig. 1 and table S5), and probably more than 2300 years if Monte Verde more accurately provides a minimum estimate of the occupation of the wider Patagonian area (*13*).

The prolonged temporal overlap between humans and megafauna in Patagonia (from around 14.6 to 13.2 ka through to the extinction events at 12.3 ka) corresponds closely in timing to the pronounced cold conditions of the ACR stadial (Fig. 1). In contrast, the tight cluster of Patagonian megafaunal extinctions occurs shortly after the start of the rapid warming phase that followed the termination of the ACR. Previous studies of southern Patagonian lake sediments have suggested that the post-ACR environment was arid and windy, with variable rainfall

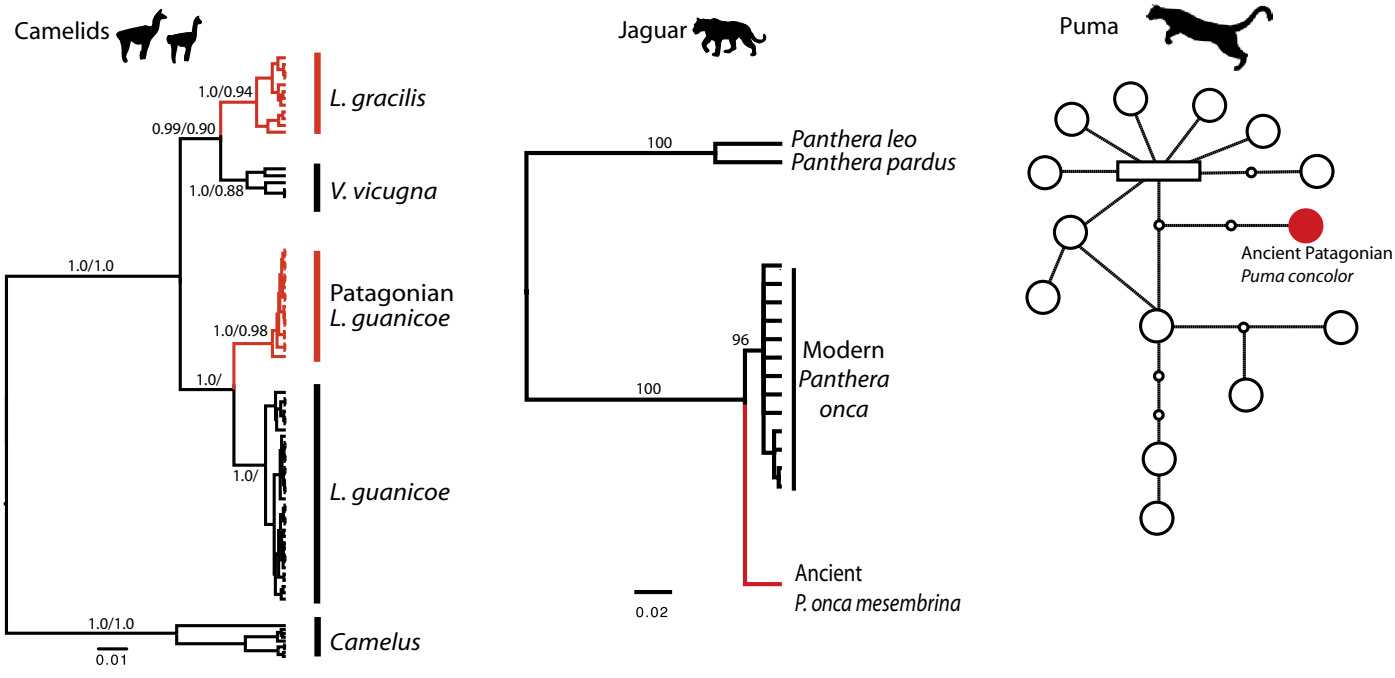

Fig. 2. Mitochondrial DNA phylogenies of Late Pleistocene Patagonian megafauna (red) relative to extant taxa (black). (**A**) Phylogenetic reconstructions of ancient and modern camelid sequences revealed two groups of camelids: *Lama gracilis* and a distinct clade of *L. guanicoe*. (**B**) Pleistocene *Panthera onca* were distinct from modern jaguars and represent the extinct subspecies *P. onca mesembrina*. (**C**) In contrast, sequences from Pleistocene *Puma concolor* are closely related to modern haplotypes. All Pleistocene samples represent haplotypes that are extinct or unsampled in modern populations.

and potential summer droughts (*11*). By 12.3 ka, when the extinctions occured, glacial ice was retreating and sufficient moisture was available to support rapid expansion of *Nothofagus* forest, consistent with the warming observed in the WAIS record (*9*).

DISCUSSION

The close association of the Patagonian megafaunal extinctions with the warming phase following the ACR agrees with recent studies suggesting that interstadials were a key driver of Holarctic megafaunal extinctions through the Late Pleistocene (*27*). However, the reversed temperature trends in North and South America during this period led to a marked inversion in the sequence of events and timing (Fig. 1). In North America, multiple megafaunal extinctions are inferred between 14.6 and 12.8 ka during the warm Greenland Interstadial 1, followed by a relative hiatus during the cold Greenland Stadial 1, before the last extinctions after 11.6 ka in the early Holocene (*27*). The pattern in southern Patagonia is temporally exactly inverted but also shows megafaunal extinctions occurred during a warming phase and a hiatus during a stadial (Fig. 1). In addition, the extinction process in North America appears considerably more protracted, at least from our data, potentially as a consequence of the hiatus during Greenland Stadial 1 in the middle of the event.

The Patagonian camelid data also appear to reflect metapopulation processes similar to those inferred from ancient DNA studies of several Late Pleistocene Northern Hemisphere megafaunal taxa, including cave bear, bison, and mammoth (*27*–*29*). Following the extinction of Patagonian *L. gracilis* and the Late Pleistocene clade, or possibly subspecies, of *L. guanicoe* around 12.3 ka, a genetically distinct northern population of *L. guanicoe* moved into the region. Early Holocene camelid bone deposits from Cerro Casa del Piedra 7 in southern Patagonia (fig. S1) provide a minimum age (10,551 ± 170 years ago) for the appearance of the replacement northern population (see the Supplementary Materials). Since the latter is ancestral to all modern guanaco stocks, this metapopulation process also appears to have been responsible for the species' survival. The observation of similar rapid continental-scale conspecific replacements in both the Holarctic and Patagonia suggests that such metapopulation structures were common (*28*, *30*–*32*), reinforcing the hypothesis that they may have evolved to provide resilience to the frequent and marked climatic shifts observed throughout the Pleistocene (*27*).

Although the Patagonian genetic record has a more limited temporal span than the Northern Hemisphere records previously reported (*27*), the higher resolution afforded by our study reveals the interplay between human impacts and climate change. The initial presence of humans in the area during the ACR stadial conditions was apparently insufficient to drive megafaunal extinctions, in direct contrast to the Blitzkrieg model, which suggests that the naïveté of megafauna to human hunting led to rapid extinction (*33*). However, human presence, in combination with the rapid advance of forests and environmental changes associated with the ensuing warming phase, appears to have led to the collapse of the megafaunal ecosystem within a few hundred years. It is unclear whether rapid increases in human population size were associated with the warming phase, although ancient DNA analyses indicate a very rapid demographic growth among the founding American populations (*34*). The new genetic data are consistent with the hypothesis that human disruption of megafaunal metapopulation processes (for example, long distance dispersal and rescue effects), along with increased hunting pressure associated with decreased habitat range, allowed local population extinctions initiated by environmental change to coalesce into a larger ecosystem-wide alteration, potentially with minimal direct signs of human hunting (*27*).

MATERIALS AND METHODS

We collected 175 Late Pleistocene and Holocene megafaunal bone and teeth fragments from museum collections in Argentina (La Plata Museum, La Plata Zoo, Instituto de Ciencias Básicas, Universidad Nacional de Cuyo, and Museo de Historia Natural de San Rafael), Chile (Centro de Estudios del Hombre Austral, Instituto de la Patagonia, and Universidad de Magallanes), Perú (Departamento Científico de Arqueología and Facultad de Ciencias Naturales y Museo), Netherlands (Zoological Museum Amsterdam), Sweden (Museum of Natural History, Malmo), and Russia (Moscow State University Zoological Museum) (tables S1 and S2). Most of the specimens were originally collected from several well-known caves in the Última Esperanza region of southern Patagonia, including Cueva del Milodón, Cueva Lago Sofía 4, and Cueva del Medio. Our sample collection also included megafauna subfossil material from caves in the Patagonia regions of Tierra del Fuego at the southernmost tip of Patagonia, the volcanic area of Pali Aike, and central Santa Cruz (fig. S1). We also sampled Holocene camelid material from these sites in Patagonia, as well as several other caves in the region (fig. S1). The Cerro Casa de Piedra site in the Santa Cruz region of Patagonia contains deposits of guanaco material spanning the early Holocene to recent times. Finally, we included several camelid samples collected from Peruvian caves to examine which lineages of camelids were present outside of Patagonia during the Pleistocene and the Holocene.

We sampled well-preserved megafaunal remains, primarily cortical limb bone and tooth roots, using a Dremel drill and disposable carborundum cutting discs. Samples of approximately 100 mg were removed and transferred to the ancient DNA laboratory.

DNA extraction and sequencing of mtDNA

We primarily carried out ancient DNA laboratory procedures at the Australian Centre for Ancient DNA (ACAD) in Adelaide, Australia, although the felid specimens had previously been examined at the Henry Wellcome Ancient Biomolecules Centre in Oxford, UK. We followed standard ancient DNA protocols for DNA extraction and polymerase chain reaction (PCR) setup in a physically isolated, clean laboratory with positive air pressure, HEPA-filtered air flow and ultraviolet(UV) lights, regular bleaching, and multiple controls for all stages of the procedures (*35*). Before extraction, we attempted to remove potential contaminants by removing the outer layer of each sample using a Dremel drill and disposable carborundum cutting discs and by exposing the sample to UV light. DNA was extracted using either phenol-chloroform (*36*) or silica-based extraction methods (*37*).

We amplified a number of phylogenetically informative mitochondrial fragments (for example, control region, cytochrome b, ATP8, and/or ND5) for each sample to confirm morphological identification (table S3). Due to the degraded nature of the samples, we amplified multiple, short, overlapping DNA fragments. We detail the PCR conditions for each group of taxa below (camelids, felids, and bear).

Camelid samples. Six control region fragments and one cytochrome b fragment were targeted for PCR using primers listed in table S3. Two sets of multiplex PCRs were performed to amplify all seven fragments (*38*). Multiplex PCRs were done in a final volume of 25 µl using 1 to 2 U of Platinum *Taq* High-Fidelity DNA Polymerase and 1× buffer (Invitrogen), rabbit serum albumin (RSA) (2 mg/ml; Sigma), 2 mM $MgSO_4$, 250 µM of each deoxynucleotide triphosphate (dNTP), 1 µM of each primer, and 1 to 4 µl of DNA. PCRs were run at 94°C for 1 min followed by 30 cycles (multiplex) or 50 cycles (singleplex) at 94°C for 15 s, 55°C for 15 s, and 68°C for 30 s, and a final elongation step at 68°C for 10 min. A no-template control and a negative extraction control were included for approximately every four samples. The second round of PCR was performed in singleplex reactions with 0.5 U of HotMaster Taq, 1× buffer (HotMaster), 200 µM of each dNTP, bovine serum albumin (BSA) (1 mg/ml), 1 µM of each primer, and a 100-fold dilution of the first-round PCR product. PCR products were visualized on a 4% tris-borate EDTA agarose gel. In rare cases in which a negative control exhibited a band, the PCR was discarded and not used for sequencing.

Felid samples. The general carnivore primers ATP8_1F/ATP8_3R were initially used to identify the species, followed by more species-specific overlapping primer pairs to amplify regions of the *Smilodon*, puma, and jaguar samples (table S3). PCR amplifications (25 µl of total volume) were done using Platinum *Taq* High-Fidelity DNA Polymerase (1.25 U; Invitrogen), 1× buffer (Invitrogen), BSA (0.2% w/v; Sigma), 2 mM $MgSO_4$, 2′-deoxynucleotide 5′-triphosphate mix (all 25 mM), 1 µl of an extract, and 1 µM each of forward and reverse primers. A 2-min activation step at 94°C was followed by 45 cycles at 94°C for 45s, an appropriate annealing temperature for 45 s, and at 68°C for 1 min and 30 s. PCR products were purified using the QIAquick system (Qiagen Ltd.). For *P. concolor*, approximately 650 base pairs (bp) of mtDNA (16S, ND5, and ATP8) were sequenced (table S3).

Bear samples. Ancient DNA extracts were initially tested using carnivore-specific primers designed to amplify a 135-bp fragment of the hypervariable region of the mtDNA control region (*39*). Additionally, for the ancient DNA extracts from which the 135-bp control region fragment was successfully amplified and sequenced, a multiplex PCR protocol (*38*) was applied, yielding a total of 135 to 364 bp of control region sequence and up to 137 bp of protein-coding ATP8 sequence and 192 bp of cytochrome b sequence (table S3). Multiplex reactions were set up in the ancient DNA facility at ACAD. Each multiplex reaction (20 µl) contained the following: 1 µl of DNA extract, 2 U of Platinum *Taq* High-Fidelity DNA Polymerase and 1× buffer (Invitrogen), RSA (1 mg/ml; Sigma), 8 mM MgSO4, 250 µM of each dNTP, and 3 µl of primer mix A or B (each containing the three primer pairs at a concentration of 1 µM). Multiplex PCR thermal cycling reactions were conducted at 94°C for 1 min, followed by 30 cycles of denaturation at 94°C for 15 s, annealing for 20 s at 55°C, and extension at 68°C for 30 s, followed by a final extension at 68°C for 10 min. The PCR master mix for the singleplex reactions contained the following: 5 µl of 1:20 dilution of multiplex PCR product, 0.25 U of Platinum *Taq* High-Fidelity DNA Polymerase and 1× buffer (Invitrogen), RSA (1 mg/ml; Sigma), $MgSO_4$, 250 µM of each dNTP, 0.75 µM forward primer, and 0.75 µM reverse primer. Singleplex PCR thermal cycling reactions were conducted at 94°C for 1 min, followed by 30 cycles of denaturation at 94°C denaturation for 15 s, annealing for 20 s at 55°C, and extension at 68°C for 30 s, followed by a final extension at 68°C for 10 min. Amplification products (from normal or singleplex PCRs described above) of the expected size were purified using ExoSAP-IT or Agencourt AMPure PCR purification kit according to the manufacturer's instructions.

We used Sanger sequencing methods with BigDye chemistry and an ABI 3130xl Genetic Analyzer or an ABI PRISM capillary DNA 377 and 310 automated sequencers (Applied Biosystems) to sequence amplicons either directly or after molecular cloning. To ensure that our results were robust, a subset was independently replicated at the Centre for GeoGenetics in Copenhagen, Denmark and the Henry Wellcome Ancient Biomolecules Centre in Oxford, UK. Below, we describe independent replication for each taxon.

Camelid samples. Independent replication for successful samples from each major taxon was performed at the Centre for GeoGenetics in Copenhagen, Denmark. Here, the samples were drilled or crushed to obtain ~280 mg (10 to 660 mg) of bone powder extracted in 0.5 M EDTA, 100 µl of proteinase K, and 0.5% SDS or *N*-lauryl-sarcosine at 37°C for 24 to 48 hours until all bone powder was digested. This was followed by centrifugation using a Millipore centrifuge at 3500*g* for 20 to 60 min and then by DNA purification using QIAquick columns and elution to 80 µl. Three samples were extracted in each session with an extraction blank. Two pools of multiplex PCR consisting of eight primers each were done as to ACAD, except 0.25 U of Taq Gold, 1× gold buffer, 4 mM $MgCl_2$, and BSA (1 mg/ml). The second round of singleplex PCR was done on a 1:20 dilution of the PCR product from the first round. Extraction blanks, and PCR controls included with every six samples, were all blank. Sanger sequencing was done at the Macrogen facilities (Korea), and sequences were assembled and aligned using Sequencher version 4.8.

Phylogenetic analysis

We estimated the best nucleotide substitution model for each mitochondrial gene for each species by selecting the model with the lowest Akaike's information criterion score using ModelTest (*40*) or other similar software. We inferred phylogenies using Bayesian methods in BEAST (*41*) and likelihood methods using PhyML (*42*, *43*), and reported Bayesian prior probabilities and maximum likelihood estimates, respectively, of highly supported nodes. For the closely related Puma haplotypes, we constructed a parsimony haplotype network using TCS (*44*). Details on the phylogenetic analysis of each taxon are included below.

Camelid samples. We aligned sequences by eye using Se-Al version 2.0a11 (http://tree.bio.ed.ac.uk/software/seal/). We reconstructed the phylogenetic relationship of camelids using Bayesian methods in BEAST (*41*) and likelihood methods using PhyML (*42*, *43*). First, we analyzed a data set that consisted of 541 nucleotides of mitochondrial sequence data (~432 bp of control region plus 107 bp of cytochrome *b*), for a total of 78 taxa, including 30 ancient Pleistocene samples, 13 ancient Holocene samples, and 35 modern camelid haplotypes, which include 5 *Camelus* haplotypes (Fig. 2). Second, we analyzed a larger sample set with 432 bp of mtDNA control region. These data included 88 ancient camelid samples and 77 modern camelid haplotypes (fig. S2). We downloaded sequence data representing modern camelid genetic diversity from GenBank (*45*, *46*).

Felid samples. Sequences were aligned by eye using Se-Al version 2.0a11 (http://tree.bio.ed.ac.uk/software/seal/). We analyzed 343 bp of control region and 143 bp of ATP8 mitochondrial sequence data from *P. onca mesembrina*. We generated a phylogeny using a UPGMA tree-building method with an HKY model of sequence evolution and

100 bootstraps (Fig. 2). We analyzed ~650 bp of 16*S*, ND5, and ATP8 of sequence data from *P. concolor* samples, and visualized their close relationship to modern haplotypes with a parsimony haplotype network (Fig. 2) using TCS (*44*).

Bear samples. We carried out phylogenetic reconstructions of Tremarctinae bears using 694 bp of mitochondrial sequence data from the *Arctotherium* sp. sample ACAD3599, along with previously sequenced Tremarctinae bears (*Tremarctos* and *Arctodus*) and previously sequenced members of Ursine bears, the sister group to Tremarctinae (*47*–*49*). We used the HKY + gamma model in BEAST 1.4.8 (*41*) to generate a Bayesian inference phylogeny. Three independent runs of 10 million Markov chain Monte Carlo generations sampled every 1000 steps were done in BEAST 1.4.8 (*41*) using a relaxed molecular clock (uncorrelated lognormal). The results of the three independent runs were combined. Results of the individual and combined runs were visualized using Tracer 1.4 (http://tree.bio.ed.ac.uk/software/tracer) to check for convergence and to ensure that all Effective Sample Size scores were >200 (suggesting sufficient sampling and run length). The first 1 million steps (10%) of each run were discarded as burn-in. The trees were annotated and visualized using FigTree 1.1.2 (http://tree.bio.ed.ac.uk/software/figtree).

^{14}C dating and calibrations

We generated 75 new date estimates of megafaunal bone and teeth using AMS ^{14}C dating, most of which were pretreated using ultrafiltration to purify the gelatin and minimize contamination [following (*50*)] at the Oxford Radiocarbon Accelerator Unit (ORAU) (table S1). The ^{14}C ages were calibrated against the SHCal13 data set (table S4) (*51*). We also calibrated previously published ^{14}C data from the large ground sloth (*M. darwinii*) (*14*, *20*, *52*), the saber-toothed cat (*S. populator*) (*22*, *53*), the giant short-faced bear (*Arctotherium* sp.) (*23*), and the South American horse (*H. saldiasi*) (*24*, *25*) [summarized in (*4*) and (*14*)]. For extinct Pleistocene megafauna, the ^{14}C ages were used to constrain the estimates of extinction ages for megafaunal taxa using the Phase model option in OxCal 4.2 (*26*) with General Outlier analysis detection (probability = 0.05) (*54*). Importantly, this approach assumes a uniform (Poisson) distribution of ^{14}C ages to estimate the calendar age boundaries (*55*); where there were less than 11 ^{14}C ages for a group, we did not use the Phase option and instead undertook individual age calibration. Using Bayes' theorem, the algorithms employed sample possible solutions with a posterior probability that is the product of the prior and likelihood probabilities; the outlier option was used to detect ages that fall outside the calibration model for each group and, if necessary, down-weight their contribution to the final age estimates. Taking into account the deposition model and the actual age measurements, the posterior probability densities quantify the likeliest age distributions. The calibrated ages are reported in table S4. To reassure ourselves that the Phase age modeling results are robust, we undertook individual calibrations of the youngest samples for each Patagonian species with greater than 11 ^{14}C dates. The ages were calibrated using SHCal13 (*50*) in OxCal 4.2 (*26*) (table S4). All calibrated ages fall within the period of rapid warming that followed the termination of the ACR, supporting the approach taken here.

We used published data sets (*56*, *57*) to estimate the arrival of humans in Patagonia, and followed their criteria for screening radiocarbon ages. We calibrated ^{14}C dates using the SHCal13 data set, as described in the previous paragraph (table S5), and estimated human arrival using the Phase model option in OxCal 4.2 (*26*) with General Outlier analysis detection (probability = 0.05) (*54*). A χ^2 test demonstrates that human arrival was statistically different to extinction in Patagonia (df = 1, T = 17.270, 5% = 3.8).

The wide range of geographic sites, depositional situations, closely spaced (and relatively young) ^{14}C ages from different species, and the statistical overlap of the Phase ages generated using OxCal 4.2 (*26*) makes it highly unlikely that the narrow temporal window of the last observations might be the result of a taphonomic or observational artifact, such as the Signor-Lipps effect (*58*). Furthermore, the density and quality of the AMS ^{14}C dates (mostly bone or teeth samples pretreated using ultrafiltration at ORAU) suggest that the chronological series are accurate. Importantly, the Signor-Lipps effect would only extend the period of apparent overlap between humans and megafauna and further confirm our observation that the megafaunal, extinctions occurred during the warming phase following the ACR stadial.

We plotted the summed probabilities of the IntCal13-calibrated (*59*) terminal AMS dates for North American *Equus caballus*, *Saiga tatarica*, *Panthera leo spelaea*, *Mammuthus primigenius*, and *Mammut americanum* from the study of Cooper *et al.* (*27*), and plotted these against time. These were the only latest Pleistocene North American megafaunal extinctions with suitably detailed ^{14}C data sets available (*27*). Timing of onset and termination of the ACR are as defined by Antarctic ice core isotopic records and change in the interhemispheric radiocarbon gradient (*7*, *9*).

SUPPLEMENTARY MATERIALS

Supplementary material for this article is available at http://advances.sciencemag.org/cgi/content/full/2/6/1501682/DC1

Supplementary Methods

Supplementary Results

fig. S1. Map of South America showing sites where genetic data were recovered.

fig. S2. Phylogeny of camelids based on 432 bp of mitochondrial control region data.

fig. S3. Mitochondrial sequence data for *Arctotherium*.

table S1. Fossil samples included in this study.

table S2. Samples for which DNA extraction failed and ^{14}C failed or was not attempted.

table S3. Primer sequences used to amplify mitochondrial genes.

table S4. Megafaunal ^{14}C ages were calibrated against the SHCal13 data set.

table S5. Published human ^{14}C ages were calibrated against the SHCal13 data set.

table S6. Results of independent replication of ancient DNA sequences.

References (*61*–*65*)

REFERENCES AND NOTES

1. J. Bird, Antiquity and migrations of the early inhabitants of Patagonia. *Geogr. Rev.* **28**, 250–275 (1938).
2. J. Clutton-Brock, *Travels and Archaeology in South Chile*, J. Bird, Ed. (University of Iowa Press, Iowa, 1988), pp. 188–195.
3. F. M. Martin, *Tafonomía de la Transición Pleistoceno-Holoceno en Fuego-Patagonia* (Ediciones de la Universidad de Magallanes, Punta Arenas, 2013).
4. A. D. Barnosky, E. L. Lindsey, Timing of Quaternary megafaunal extinction in South America in relation to human arrival and climate change. *Quat. Int.* **217**, 10–29 (2010).
5. N. A. Villavicencio, E. L. Lindsey, F. M. Martin, L. A. Borrero, P. I. Moreno, C. R. Marshall, A. D. Barnosky, Combination of humans, climate, and vegetation change triggered Late Quaternary megafauna extinction in the Última Esperanza region, southern Patagonia, Chile. *Ecography* **38**, 125–140 (2015).
6. F. Lamy, J. Kaiser, U. Ninnemann, D. Hebbeln, H. W. Arz, J. Stoner, Antarctic timing of surface water changes off Chile and Patagonian ice sheet response. *Science* **304**, 1959–1962 (2004).
7. J. B. Pedro, H. C. Bostock, C. M. Bitz, F. He, M. J. Vandergoes, E. J. Steig, B. M. Chase, C. E. Krause, S. O. Rasmussen, B. R. Markle, G. Cortese, The spatial extent and dynamics of the Antarctic Cold Reversal. *Nat. Geosci.* **9**, 51–55 (2015).
8. C. J. Fogwill, C. S. M. Turney, D. K. Hutchinson, A. S. Taschetto, M. H. England, Obliquity control on southern hemisphere climate during the last glacial. *Sci. Rep.* **5**, 11673 (2015).

9. WAIS Divide Project Members, Precise interpolar phasing of abrupt climate change during the last ice age. *Nature* **520**, 661–665 (2015).
10. D. E. Sugden, M. J. Bentley, C. J. Fogwill, N. R. J. Hulton, R. D. McCulloch, R. S. Purves, Late-glacial glacier events in southernmost South America: A blend of 'northern' and 'southern' hemispheric climatic signals? *Geogr. Ann.* **87**, 273–288 (2005).
11. P. I. Moreno, M. R. Kaplan, J. P. François, R. Villa-Martínez, C. M. Moy, C. R. Stern, P. W. Kubik, Renewed glacial activity during the Antarctic cold reversal and persistence of cold conditions until 11.5 ka in southwestern Patagonia. *Geology* **37**, 375–378 (2009).
12. T. D. Dillehay, C. Ramírez, M. Pino, M. B. Collins, J. Rossen, J. D. Pino-Navarro, Monte Verde: Seaweed, food, medicine, and the peopling of South America. *Science* **320**, 784–786 (2008).
13. T. D. Dillehay, C. Ocampo, J. Saavedra, A. O. Sawakuchi, R. M. Vega, M. Pino, M. B. Collins, L. Scott Cummings, I. Arregui, X. S. Villagran, G. A. Hartmann, M. Mella, A. González, G. Dix, New archaeological evidence for an early human presence at Monte Verde, Chile. *PLOS One* **10**, e0141923 (2015).
14. J. Steele, G. Politis, AMS ^{14}C dating of early human occupation of southern South America. *J. Archaeol. Sci.* **36**, 419–429 (2009).
15. F. M. Martin, L. A. Borrero, Climate change, availability of territory, and Late Pleistocene human exploration of Ultima Esperanza, South Chile. *Quat. Int.*, 10.1016/j.quaint.2015.06.023 (2015).
16. L. A. Borrero, The prehistoric exploration and colonization of Fuego-Patagonia. *J. World Prehist.* **13**, 321–355 (1999).
17. J. Weinstock, B. Shapiro, A. Prieto, J. C. Marín, B. A. González, M. T. P. Gilbert, E. Willerslev, The Late Pleistocene distribution of vicuñas (*Vicugna vicugna*) and the "extinction" of the gracile llama ("*Lama gracilis*"): New molecular data. *Quat. Sci. Rev.* **28**, 1369–1373 (2009).
18. A. Cabrera, Los yaguares vivientes y extinguidos de la America Austral. *Notas preliminares del Museuo de la Plata* **2**, 34–50 (1934).
19. M. Höss, A. Dilling, A. Currant, S. Pääbo, Molecular phylogeny of the extinct ground sloth *Mylodon darwinii*. *Proc. Natl. Acad. Sci. U.S.A.* **93**, 181–185 (1996).
20. V. Markgraf, Late Pleistocene faunal extinctions in southern Patagonia. *Science* **228**, 1110–1112 (1985).
21. J. Canto, Posible presencia de una variedad de *Smilodon* en el Pleistoceno Tardío en Magallanes. *An. Inst. Patagonia.* **20**, 96 (1991).
22. R. Barnett, I. Barnes, M. J. Phillips, L. D. Martin, C. R. Harington, J. A. Leonard, A. Cooper, Evolution of the extinct Sabretooths and the American cheetah-like cat. *Curr. Biol.* **15**, R589–R590 (2005).
23. F. J. Prevosti, F. M. Martin, Paleoecology of the mammalian predator guild of Southern Patagonia during the latest Pleistocene: Ecomorphology, stable isotopes, and taphonomy. *Quat. Int.* **305**, 74–84 (2013).
24. M. T. Alberdi, A. N. Menegaz, J. L. Prado, Formas terminales de *Hippidion* (Mammalia, Perissodactyla) de los yacimientos del Pleistoceno Tardío-Holoceno de la Patagonia (Argentina y Chile). *Estud. Geol.* **43**, 107–115 (1987).
25. L. Orlando, J. L. Metcalf, M. T. Alberdi, M. Telles-Antunes, D. Bonjean, M. Otte, F. Martin, V. Eisenmann, M. Mashkour, F. Morello, J. L. Prado, R. Salas-Gismondi, B. J. Shockey, P. J. Wrinn, S. K. Vasil'ev, N. D. Ovodov, M. I. Cherry, B. Hopwood, D. Male, J. J. Austin, C. Hänni, A. Cooper, Revising the recent evolutionary history of equids using ancient DNA. *Proc. Natl. Acad. Sci. U.S.A.* **106**, 21754–21759 (2009).
26. C. B. Ramsey, S. Lee, Recent and planned developments of the program OxCal. *Radiocarbon* **55**, 720–730 (2013).
27. A. Cooper, C. Turney, K. A. Hughen, B. W. Brook, H. G. McDonald, C. J. A. Bradshaw, Abrupt warming events drove Late Pleistocene Holarctic megafaunal turnover. *Science* **349**, 602–606 (2015).
28. D. H. Mann, P. Groves, R. E. Reanier, B. V. Gaglioti, M. L. Kunz, B. Shapiro, Life and extinction of megafauna in the ice-age Arctic. *Proc. Natl. Acad. Sci. U.S.A.* **112**, 14301–14306 (2015).
29. D. Nogués-Bravo, J. Rodríguez, J. Hortal, P. Batra, M. B. Araújo, Climate change, humans, and the extinction of the woolly mammoth. *PLOS Biol.* **6**, e79 (2008).
30. A. L. Cione, E. P. Tonni, L. Soibelzon, The broken Zig-Zag: Late Cenozoic large mammal and tortoise extinction in South America. *Rev. Mus. Argentino Cienc. Nat. N. S.* **5**, 1–19 (2003).
31. A. L. Cione, E. P. Tonni, L. Soibelzon, *Vertebrate Paleobiology and Paleoanthropology Series,* G. Haynes, Ed. (Springer, Dordrecht, Netherlands, 2009), pp. 125–144.
32. M. S. Lima-Ribeiro, D. Nogués-Bravo, L. C. Terribile, P. Batra, J. A. F. Diniz-Filho, Climate and humans set the place and time of Proboscidean extinction in late Quaternary of South America. *Palaeogeogr. Palaeoclimatol. Palaeoecol.* **392**, 546–556 (2013).
33. P. S. Martin, The discovery of America. *Science* **179**, 969–974 (1973).
34. B. Llamas, L. Fehren-Schmitz, G. Valverde, J. Soubrier, S. Mallick, N. Rohland, S. Nordenfelt, C. Valdiosera, S. M. Richards, A. Rohrlach, M. I. B. Romero, I. F. Espinoza, E. T. Cagigao, L. W. Jiménez, K. Makowski, I. S. L. Reyna, J. M. Lory, J. A. B. Torrez, M. A. Rivera, R. L. Burger, M. C. Ceruti, J. Reinhard, R. S. Wells, G. Politis, C. M. Santoro, V. G. Standen, C. Smith, D. Reich, S. Y. W. Ho, Alan Cooper, W. Haak, Ancient mitochondrial DNA provides high-resolution timescale of the peopling of the Americas. *Sci. Adv.* **2**, e1501385 (2016).
35. E. Willerslev, A. Cooper, Review paper. Ancient DNA. *Proc. Biol. Sci.* **272**, 3–16 (2005).
36. J. Weinstock, E. Willerslev, A. Sher, W. Tong, S. Y. W. Ho, D. Rubenstein, J. Storer, J. Burns, L. Martin, C. Bravi, A. Prieto, D. Froese, E. Scott, L. Xulong, A. Cooper, Evolution, systematics, and phylogeography of Pleistocene horses in the New World: A molecular perspective. *PLOS Biol.* **3**, e241 (2005).
37. P. Brotherton, W. Haak, J. Templeton, G. Brandt, J. Soubrier, C. J. Adler, S. M. Richards, C. Der Sarkissian, R. Ganslmeier, S. Friederich, V. Dresely, M. van Oven, R. Kenyon, M. B. Van der Hoek, J. Korlach, K. Luong, S. Y. W. Ho, L. Quintana-Murci, D. M. Behar, H. Meller, K. W. Alt, A. Cooper, S. Adhikarla, A. K. G. Prasad, R. Pitchappan, A. V. Santhakumari, E. Balanovska, O. Balanovsky, J. Bertranpetit, D. Comas, B. Martínez-Cruz, M. Melé, A. C. Clarke, E. A. Matisoo-Smith, M. C. Dulik, J. B. Gaieski, A. C. Owings, T. G. Schurr, M. G. Vilar, A. Hobbs, H. Soodyall, A. Javed, L. Parida, D. E. Platt, A. K. Royyuru, L. Jin, S. Li, M. E. Kaplan, N. C. Merchant, R. J. Mitchell, C. Renfrew, D. R. Lacerda, F. R. Santos, D. F. S. Hernanz, R. S. Wells, P. Swamikrishnan, C. Tyler-Smith, P. P. Vieira, J. S. Ziegle; The Genographic Consortium, Neolithic mitochondrial haplogroup H genomes and the genetic origins of Europeans. *Nat. Commun.* **4**, 1764 (2013).
38. H. Römpler, P. H. Dear, J. Krause, M. Meyer, N. Rohland, T. Schöneberg, H. Spriggs, M. Stiller, M. Hofreiter, Multiplex amplification of ancient DNA. *Nat. Protoc.* **1**, 720–728 (2006).
39. C. Hänni, V. Laudet, D. Stehelin, P. Taberlet, Tracking the origins of the cave bear (*Ursus spelaeus*) by mitochondrial DNA sequencing. *Proc. Natl. Acad. Sci. U.S.A.* **91**, 12336–12340 (1994).
40. D. Posada, K. A. Crandall, MODELTEST: Testing the model of DNA substitution. *Bioinformatics* **14**, 817–818 (1998).
41. A. J. Drummond, A. Rambaut, BEAST: Bayesian evolutionary analysis by sampling trees. *BMC Evol. Biol.* **7**, 214 (2007).
42. S. Guindon, O. Gascuel, A simple, fast, and accurate algorithm to estimate large phylogenies by maximum likelihood. *Syst. Biol.* **52**, 696–704 (2003).
43. S. Guindon, F. Lethiec, P. Duroux, O. Gascuel, PHYML Online—A web server for fast maximum likelihood-based phylogenetic inference. *Nucleic Acids Res.* **33**, W557–W559 (2005).
44. M. Clement, D. Posada, K. A. Crandall, TCS: A computer program to estimate gene geneologies. *Mol. Ecol.* **9**, 1657–1659 (2000).
45. J. C. Marín, A. E. Spotorno, B. A. González, C. Bonacic, J. C. Wheeler, C. S. Casey, M. W. Bruford, R. E. Palma, E. Poulin, Mitochondrial DNA variation and systematics of the guanaco (*Lama guanicoe*, Artiodactyla: Camelidae). *J. Mammal.* **89**, 269–281 (2008).
46. J. C. Marín, B. Zapata, B. A. González, C. Bonacic, J. C. Wheeler, C. Casey, M. W. Bruford, R. Eduardo Palma, E. Poulin, M. A. Alliende, A. E. Spotorno, Systematics, taxonomy and domestication of alpaca and llama: New chromosomal and molecular evidence. *Rev. Chil. Hist. Nat.* **80**, 121–140 (2007).
47. I. Delisle, C. Strobeck, Conserved primers for rapid sequencing of the complete mitochondrial genome from carnivores, applied to three species of bears. *Mol. Biol. Evol.* **19**, 357–361 (2002).
48. J. Krause, T. Unger, A. Noçon, A.-S. Malaspinas, S.-O. Kolokotronis, M. Stiller, L. Soibelzon, H. Spriggs, P. H. Dear, A. W. Briggs, S. C. E. Bray, S. J. O'Brien, G. Rabeder, P. Matheus, A. Cooper, M. Slatkin, S. Pääbo, M. Hofreiter, Mitochondrial genomes reveal an explosive radiation of extinct and extant bears near the Miocene-Pliocene boundary. *BMC Evol. Biol.* **8**, 220 (2008).
49. R. Peng, B. Zeng, X. Meng, B. Yue, Z. Zhang, F. Zou, The complete mitochondrial genome and phylogenetic analysis of the giant panda (*Ailuropoda melanoleuca*). *Gene* **397**, 76–83 (2007).
50. C. B. Ramsey, T. Higham, A. Bowles, R. Hedges, Improvements to the pretreatment of bone at Oxford. *Radiocarbon* **46**, 155–163 (2004).
51. A. G. Hogg, Q. Hua, P.G. Blackwell, M. Niu, C. E. Buck, T. P. Guilderson, S. R. Zimmerman, SHCal13 Southern Hemisphere calibration, 0–50,000 years cal BP. *Radiocarbon* **55**, 1889–1903 (2013).
52. A. Prieto, Cazadores tempranos y tardíos en cueva del lago sofía 1. *An. Inst. Patagonia* **20**, 75 (1991).
53. A. Prieto, R. Labarca, V. Sierpe, New evidence of the sabertooth cat *Smilodon* (Carnivora: Machairodontinae) in the late Pleistocene of southern Chilean Patagonia. *Rev. Chil. Hist. Nat.* **83**, 299–307 (2010).
54. C. B. Ramsey, Dealing with outliers and offsets in radiocarbon dating. *Radiocarbon* **51**, 1023–1045 (2009).
55. C. E. Buck, C. D. Litton, A. F. M. Smith, Calibration of radiocarbon results pertaining to related archaeological events. *J. Archaeol. Sci.* **19**, 497–512 (1992).
56. F. M. Martín, Human–carnivore interaction at the end of the pleistocene in Southern Patagonia, Chile. *J. Taphon.* **10**, 561–574 (2012).
57. L. Prates, G. Politis, J. Steele, Radiocarbon chronology of the early human occupation of Argentina. *Quat. Int.* **301**, 104–122 (2013).
58. F. Saltré, B. W. Brook, M. Rodríguez-Rey, A. Cooper, C. N. Johnson, C. S. M. Turney, C. J. A. Bradshaw, Uncertainties in dating constrain model choice for inferring extinction time from fossil records. *Quat. Sci. Rev.* **112**, 128–137 (2015).

59. P. J. Reimer, E. Bard, A. Bayliss, J. W. Beck, P. G. Blackwell, C. B. Ramsey, C. E. Buck, H. Cheng, R. L. Edwards, M. Friedrich, P. M. Grootes, T. P. Guilderson, H. Haflidason, I. Hajdas, C. Hatté, T. J. Heaton, D. L. Hoffmann, A. G. Hogg, K. A. Hughen, K. F. Kaiser, B. Kromer, S. W. Manning, M. Niu, R. W. Reimer, D. A. Richards, E. M. Scott, J. R. Southon, R. A. Staff, C. S. M. Turney, J. van der Plicht, IntCal13 and marine13 radiocarbon age calibration curves 0–50,000 years Cal BP. *Radiocarbon* **55**, 1869–1887 (2013).
60. S. O. Rasmussen, M. Bigler, S. P. Blockley, T. Blunier, S. L. Buchardt, H. B. Clausen, I. Cvijanovic, D. Dahl-Jensen, S. J. Johnsen, H. Fischer, V. Gkinis, M. Guillevic, W. Z. Hoek, J. J. Lowe, J. B. Pedro, T. Popp, I. K. Seierstad, J. P. Steffensen, A. M. Svensson, P. Vallelonga, B. M. Vinther, M. J. C. Walker, J. J. Wheatley, M. Winstrup, A stratigraphic framework for abrupt climatic changes during the last glacial period based on three synchronized Greenland ice-core records: Refining and extending the INTIMATE event stratigraphy. *Quat. Sci. Rev.* **106**, 14–28 (2014).
61. L. A. Borrero, Pleistocene extinctions in South America, in *Quaternary of South America and Antarctic Peninsula* (Balkema, Leiden, Netherlands, 1984) vol. 2, pp. 115–125.
62. H. Gervais, F. Ameghino, Los Mamíferos Fósiles de la América Meridional (Librairie F. Savy, Paris, 1880).
63. J. Cajal, E. P. Tonni, V. Tartarini, The extinction of some South American camelids: the case of *Lama* (*Vicugna*) *gracilis*. *Mastozool. Neotrop.* **17**, 129–134 (2010).
64. I. Cartajena, P. López, I. Martínez, New camelid (Artiodactyla: Camelidae) record from the late Pleistocene of Calama (Second Region, Chile): A morphological and morphometric discussion. *Rev. Mex. Cienc. Geol.* **27**, 197–212 (2010).
65. L. H. Pomi, F. J. Prevosti, Sobre el status sistemático de Felis longifrons Burmeister, 1866 (Carnivora: Felidae). *Ameghiniana* **42**, 489–494 (2005).

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